

We claim:

1. A composition comprising a substantially purified thermostable Gux1 peptide, said Gux1 peptide comprising a catalytic domain GH48, a carbohydrate binding domain (CBD) type III, and a carbohydrate binding domain (CBD) type II.

2. The composition of claim 1 wherein the Gux1 peptide is further defined as comprising a linker and a signal peptide.

3. The composition of claim 1 or 2 wherein the GH48 catalytic domain of the Gux1 peptide is further defined as having a length of about 637 to about 643 amino acids.

4. The composition of claim 1, 2, or 3 wherein the carbohydrate binding domain (CBD) type III of the Gux1 peptide is further defined as having a length of about 150 to about 156 amino acids.

5. The composition of claim 1, 2, 3, or 4 wherein the carbohydrate binding domain (CBD) type II of the Gux1 peptide is further defined as having a length of about 95 amino acids to about 105 amino acids in length.

6. The composition of claim 3 wherein the GH48 catalytic domain is further defined as the sequence of SEQ ID NO: 5.

7. The composition of claim 4 wherein the carbohydrate binding domain (CBD) type III is further defined as the sequence of SEQ ID NO: 4.

8. The composition of claim 6 wherein the carbohydrate binding domain (CBD) type II is further defined as the sequence of SEQ ID NO: 7.

9. The composition of claim 1 further defined as comprising a sequence of SEQ ID NO: 4, SEQ ID NO: 5, and SEQ ID NO: 7.

10. A thermal tolerant Gux1 peptide having a sequence of SEQ ID NO: 1.

11. The Gux1 peptide of claim 10 further defined as having a sequence of SEQ ID NO: 2.

12. An industrial mixture suitable for degrading cellulose, such mixture comprising the Gux1 polypeptide of claim 1.

13. The industrial mixture of claim 12 further defined as comprising a detergent.

14. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence having at least 90% sequence identity to the nucleic acid sequence encoding an amino acid sequence of SEQ ID NO: 5.

15. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence having at least 80% sequence identity to the nucleic acid sequence encoding an amino acid sequence of SEQ ID NO: 5.

16. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence having at least 70% sequence identity to the nucleic acid sequence encoding an amino acid sequence of SEQ ID NO: 5.

17. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence having at least 90% sequence identity to the nucleic acid sequence encoding the amino acid sequence of SEQ ID NO: 7.

18. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence having at least 90% sequence identity to the nucleic acid sequence encoding the amino acid sequence of SEQ ID NO: 4.

19. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence having at least 90% sequence identity to the nucleic acid sequence encoding the amino acid sequence of SEQ ID NO: 6.

20. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence having at least 90% sequence identity to the nucleic acid sequence encoding the amino acid sequence of SEQ ID NO: 1.

5 21. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence having at least 90% identity to the nucleic acid sequence of SEQ ID NO: 2.

22. The composition of claim 1 wherein the Gux1 is further defined as comprising a nucleic acid sequence encoding a heterologous protein in frame with the Gux1 peptide of claim 1.

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23. The composition of claim 22 wherein the heterologous protein in frame with the Gux1 peptide of claim 1 is further defined as a peptide tag.

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24. The composition of claim 23 wherein the peptide tag is 6-His, thioredoxin, hemagglutinin, GST, or OmpA signal sequence tag.

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24.25 The composition of claim 22 wherein the heterologous protein is a substrate targeting moiety.

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20 25. The composition of claim 13 wherein the nucleotide sequence encoding the Gux1 is operably linked to a transcriptional or translational regulatory sequence.

27 26. The composition of claim ²⁶25, wherein the transcriptional or translational regulatory sequence comprises a transcriptional promoter or enhancer.

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27 27. An isolated polypeptide molecule comprising:

- a) a sequence of SEQ ID NO: 4;
- b) a sequence of SEQ ID NO: 5;
- c) a sequence of SEQ ID NO: 6;
- 30 d) a sequence of SEQ ID NO: 7;
- e) a sequence of SEQ ID NO: 1; or

f) an amino acid sequence having at least 70% sequence identity with the amino acid sequence of a), b), c), d), or e).

28. The polypeptide molecule of claim ²⁸27, having at least 90% sequence identity with the amino acid sequence of a), b), c), d), or e).

29. A fusion protein comprising the polypeptide of claim ²⁸27 and a heterologous peptide.

30. The fusion protein of claim ³⁰29, wherein the heterologous peptide is a substrate targeting moiety.

31. The fusion protein of claim ³⁰29, wherein the heterologous peptide is a peptide tag.

32. The fusion protein of claim 31, wherein the peptide tag is 6-His, thioredoxin, hemagglutinin, GST, or OmpA signal sequence tag.

33. The fusion protein of claim ³⁰29, wherein the heterologous peptide is an agent that promotes polypeptide oligomerization.

34. The fusion protein of claim ³⁰29, wherein the agent is a leucine zipper.

35. A cellulase-substrate complex comprising the isolated polypeptide molecule of claim 27 bound to cellulose.

36. A vector comprising the polynucleotide molecule that encodes a polypeptide of claim 27.

37. A host cell genetically engineered to express the polypeptide molecule of claim 27.

38. A host cell genetically engineered to express the polynucleotide molecule of claim 27.

39. The host cell of claim 37 or 38, wherein the host cell is a plant cell.

Rule 1.126
41 40.
The host cell of claim 37 or 38, wherein the host cell is a fungi.

42 41. The host cell of claim 37 or 38, wherein the host cell is a bacterial cell.

5 43 42. The host cell of claim 37 or 38, wherein the host cell is a bacterial cell.

44 43. A composition comprising the polypeptide molecule of claim 27 and a carrier.

45 44. A composition comprising the polypeptide molecule of claim 28 and a carrier.

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46 45. An isolated antibody that specifically binds to the polypeptide molecule of claim 27.

47 46. The antibody of claim 45, wherein the antibody is a polyclonal antibody.

48 47. The antibody of claim 45, wherein the antibody is a monoclonal antibody.

49 48. A method for producing Gux1 polypeptide, the method comprising:
incubating a host cell genetically engineered to express the polynucleotide molecule of claim

50 27.

51 49. The method of claim 48, further comprising the step of:
isolating the Gux1 polypeptide from the incubated host cells.

52 50. The method of claim 48, wherein the host cell is a plant cell.

53 51. The method of claim 48, wherein the host cell is a bacterial cell.

54 52. The method of claim 48, wherein the host cell is genetically engineered to express a
selectable marker.

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53 53. The method of claim 48, wherein the host cell further comprises a polynucleotide molecule encoding one or more polypeptide molecules selected from the glycoside hydrolase family of proteins.

54 54. The method of claim 53, wherein the glycoside hydrolase is a thermostable glycoside hydrolase.

55 55. A set of amplification primers for amplification of a polynucleotide molecule encoding Gux1, comprising:

10 two or more sequences comprising 9 or more contiguous nucleic acids derived from the polypeptide molecule of claim 27.

56 56. A probe for hybridizing to a polynucleotide encoding Gux1, comprising:
a sequence of 9 or more contiguous nucleic acids derived from the polypeptide molecule of claim 27.

57 57. An assay method for the detection of a polynucleotide encoding Gux1, comprising:
amplifying a nucleic acid sequence with a set of amplification primers comprising two or more sequences of 9 or more contiguous nucleic acids derived from the polypeptide molecule of claim 27; and
correlating the amplified nucleic acid sequence with detected polypeptide encoding Gux1.

58 58. A method for assessing the carbohydrate degradation activity of Gux1 comprising:
analyzing a carbohydrate degradation in the presence of Gux1 and a carbohydrate degradation
25 in the absence of Gux1 on a substrate; and
comparing the carbohydrate degradation in the presence of Gux1 with the carbohydrate degradation in the absence of Gux1.

59 59. A method for assessing the carbohydrate degradation activity of Gux1 in the presence of an
30 agent of interest comprising:
analyzing a carbohydrate degradation in the presence of Gux1 and a carbohydrate degradation in the presence of Gux1 and the agent of interest on a substrate exposed; and

comparing the carbohydrate degradation in the Gux1 treated substrate with the carbohydrate degradation in the Gux1 treated substrate in the presence of the agent of interest.

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60. The method of claim 59, wherein an increase in carbohydrate degradation activity in the presence of the agent of interest demonstrates stimulation of Gux1 activity and wherein a decrease in carbohydrate degradation activity demonstrates inhibition of Gux1 activity.

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61. The method of claim 58, wherein the carbohydrate is cellulose.

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10 62. The method of claim 58 wherein the agent of interest is an antibody.

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63. A method for reducing cellulose in a starting material, the method comprising:
administering to the starting material an effective amount of a polypeptide molecule of claim 27.

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64. The method of claim 63, further comprising administering a second polypeptide molecule selected from the glycoside hydrolase family of proteins.

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65. The method of claim 63, wherein the polypeptide molecule of claim 27 is thermostable.

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66. The method of claim 63, wherein the starting material is agricultural biomass.

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67. The method of claim 63, wherein the starting material is municipal solid waste.

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